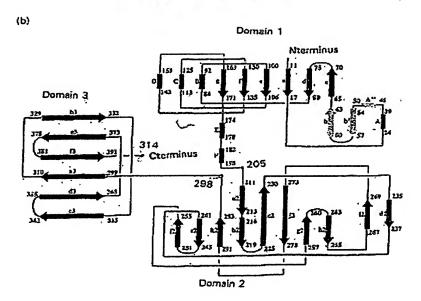
WO 2004/805506 PCT/EPZI/OJ/807068

Domain 2



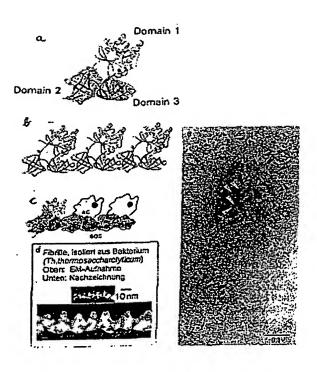
[Key to Figure 1:]

Figur = Figure

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Figur 2



[Key to Figure 2:]

Figur = Figure

Fibrille, isoliert aus Bakterium = Fibrilla, isolated from the bacterium

Oben: EM-Aufnahme = Above: EM image

Unten: Nachzeichnung = Below: Copy

```
PCT/RP2003/007068
      WO 2004/005506
                                         JI15
    Fig. 3
    (a1)
    Veldor pBGFP (Clontech):
                             STOP
   GAC GAG CTO TAC AAG TAA AGC GGC CGC GAC TCT AGA ATT CCA
CTG CTC GAC ATG TTC ATT TCG CCG GCG CTG AGA TCT TAA GGT
   BsrGI-Schnittstalle:
                                         EcoRI-Schnittstelle:
                                                AATTC
           GTACA
                                            CTTAA
      ACATG
   Synthetisch hergestelltes Oligonukleotid zur Einklonierung des His-Tags in den Vektor:
                                                         BarGI
   G TAC AAG CIT CAT CAC CAT CAC CAT CAC TAA CTG TAC AAG TAAG
TTC GAA GTA GTG GTA GTG GTA GTG ATT GAC ATG TTC ATTCTTAA
     Tyr-Lys-Leu-His-His-His-His-His-STOP-
   Ergebois: pEGFP(His)
   (a2)
   Vektor pEGFP(His):
   GCC TGC AGG -8- ACC ATG GTG
   CGG ACG TCC -4- TGG TAC CAC
     Pstl-Schnittstelle:
                                        NeoI-Schnittstelle:
                                            CATGG
     CTGCA
          ACGTC
  Fusionastellen zum EF-Tu-Gen:
                                                                     Start EGFP
                       Start EF-Tu
                                                      HindIII
                                                                 NCOI
        PatT
  ACT AGC TGC AGC ATG TCT AAA - E- CTG GGC AAG CTT ACC ATG GTG
  TGA TCG ACG TCG TAC AGA TIT -t- GAC CCG TTC GAA TGG TAC CAC
  Thr-Ser-Cys-Ser-Met-Ser-Lys-----Leu-Cly-Lys-Leu- Thr-Met-Val
[Key to Figure 3:]
Vektor = Vector
```

Schnittstelle = Interface

Synthetisch hergestelltes Oligonukleotid zur Einklonierung des His-Tags in den Vektor =

Synthetically produced oligonucleotide for cloning the His tag in the vector

Ergebnis = Result

Fusionsstellen zum EF-Tu-Gen = Fusion points in the EF-Tu gene

PC

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(a3)

Pusionsstellen zur Domäne 3:

5' PSTI CYS HindIU MODI 3'
ACT AGC TGC AGC GCT AAG CCG -1- CTG GGC TGC AAG CTT ACC ATG GTG
TGA TCG ACG TGG CGA TTC GGC -1- GAC CCG ACG TTC GAA TGG TAC CAC
3''
Thr-Sor-Cys-Ser-Als-Lys-Pro----Leu-Gly-Cys-Lys-Leu-Thr-Met-Val

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[Key to 4/15:]

Fusionsstellen zum Domäne 3 = Fusion points in domain 3

PCT/EP2003/007068 WO 2004/005506 5/15 Sequent des Konstrukts EP-Tu-GPP-His im Vektor pBGFP (Clontech) (52Q ID NO:1)

CATTCATTAB

(PT)

ATCTCTANG F TCTGACGGT G GGGAGGAAA F GGCGACTGAC TGCGACTGAC F TACGACTTCC GGAAATCCTG G GAAGAGTTG CGAAGGCCGT CTAAGCCGGG

AGGGGAGGN GCTGTTCACC GGGGTGGTGC CCATCCTGGT CGAGCTGGAC GGCGACGTAR ACGGCCACAR

Sequenz des Konstrukts EF-Tu-GFP-His im Vektor pEGFP = Sequence of the construct EF-Tu-GFP-His in the vector pEGFP

Vektor = Vector

[Key to 5/15:]

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nra-rad:

CPTCATCACC ATCACCATCA CTAACTOTAC AAGTAACATTEETE

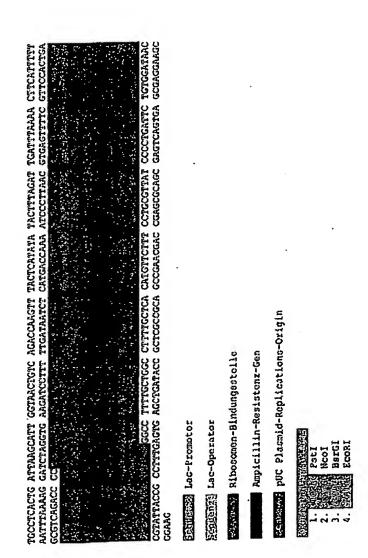
FP-Vektor

[Key to 6/15:]

Vektor = Vector

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[Key to 7/15:]

Sequenz: Lac-Promotor = Sequence: Lac-promoter

Ribosomen-Bindungsstelle = Ribosome binding site

Ampicillin-Resistenz-Gen = Ampicillin resistance gene

Klonierungsstellen = Cloning sites

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The sequence contains four silent mutations () that are clearly present according to sequence analysis:

- (1) To be: TAT, Is: TA -> Tyr; codon usage (entire E. coli genome) is changed from 16.2 to 12.2
- (2) To be: TAC, Is: TA -> Tyr; codon usage (entire E. coli genome) is changed from 12.2 to 16.2
- (3) To be: GCA, Is: GC -> Ala; codon usage (entire E. coli genome) is changed from 20.1 to 33.6
- (4) To be: ATT, Is: AT -> IIe; codon usage (entire E. coli genome) is changed from 30.3 to 25.1 (frequency per thousand)

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Sequenz des Konstrukts Domäne 3 von EF-Tu-GFP-His im Vektor pEGFP (Clontech) (SEQ ID NO:2)

DEGEP-Vextor:

CATCCETTERNE

Domäne 3 von EF-Tu:

GCTAAGOOGG GCACCATCAA GCCGCACACC ANGTICGANI CIGAAGIGIA CATICIGIC AAAGAIGAAG GCGGCCGICA IACTAAGAC TACCALGGAA CTGCCGGAAG GCGTAGAGAI GTAAIGGCI ACCGICGCAA TCAAAAAGGCI TGITACCCIG AICCACCCCGA ICGCGAIGGA CACGGICTG CGITAGAGAI GCTAAIGGC GGCGAAAAAAGGC TGITAGCGGG GCGITGIAGC AICCACAAA ICCCIGAAGG CGGCGAAAC GITGGCGGG GCGITGIAGC TAAAGIICIG GGCIGC

peger-vektor: Aagctta@

FD.

CCTGAGCAAA GACCCCAACG TGGACGAGCEEEERGAAG GCAGAAGAAC GACCACTACC AGCAGAACAC ACCECCAAGC CCACATGAAG TTCAAGGACG ACGCCAACTA TTCAAGGAGG ACGCCCACAA GCCGACGTAA CATCTGCACC GCTACCCCCA TGGCCGACAA GGGCATCGAC ACTCTCGGCA CACCATCTTC TCGAGCTGAA GICTATATCA CCAGCTCCCC CCCAGTCCGC CGAGCTGGAC CCCTGAAGTT TGCTTCAGCC CGCCGGGATC TCCAGGAGCG GTGAACCGCA GGCANGCTGA CGGCGTGCAG CAGCCACAAC TACCTGAGCA CCATCCTGGT ACGCCAGCGT GNAGGCTACG ANCATCGAGG TCGTGACCGC ACANCTACAA GGGGTGGTGC TGCCACCTAC CCCTGACCTA CGACACCCTG CGACAACCAC CTGGCCCACC CTGGTGACCA TCTTCAAGTC CGCCATGCCC AAGCTGGAGT GATCCGCCAC CTGCTGGAGT GCTGTTCACC GCGAGGGCGA TGCTGCTGCC AGTTCGAGGG GCCGAGGTGA CCTGGGGCAC TCACATGGTC TGAACTTCAA GACGCCCCC TCCGGCGAGG MEGTGAGCA AGGGCGAGCA AGNAGCGCGA CCCCATCGGC ACGCCAACAT GGCATCAAGG GTTCAGCGTG TGCCGTGCC CAGCACGACT CAAGACCCGC

His-Tag:

CTTCATCACC ATCACCATCA CTAACTGTAC AAGTAAGREEGE

[Key to 9/15:]

Sequenz des Konstrukts Domäne 3 von EF-Tu-GFP-His im Vektor pEGFP (Clontech) (SEQ ID

No: 2) (b2) = Sequence of the domain-3 construct of EF-Tu-GFP-His in the vector pEGFP (Clontech) (SEQ ID No: 2) (b2)

Vektor = Vector

Domäne = Domain

PEGEP-Vektor:

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CTGAGATAGG TTGTCTGTAA ACTATGCGGC TACCGCATCA CGTCAGGTGG CTCATGAGAC CITCATTITI GTTCCACTGA CCCTCCCGTA TCGTAGTTAT CTACACGACG GGGAGTCAGG CAACTATGGA TGAACGAAAT AGACAGATCG TGATTTAAAA GGCTGGCTTA GITTCTIAGA CGGTCACAGC AAGGAGAAAA TATGTATCCG CGGGTGTCGG ACAGATGCCT TACTITAGAT CATGACCAAA ATCCCTTAAC ATAGGCCTAC CTCCCGGAGA GATAATAATG TACATTCAAA TACTCATATA TTTTTCTABA CGGGTGTTGG GAAATACCGC ACACATGCAG TTAATGTCAT GRAPANGGAN GAGT AAAACCTCTG TATGCGGTGT GCCGCGTCAG TTTTTTAGG TATITGITTA AGACCAAGTT CTTGTCTGGT GATGACGGTG GGTAACTGTC GATACGCCTA GCGGAACCCC CCATTACCAA GAGTGCACCA CAATAATAT PATITIADANG GATCIAGGIG PAGAICCILI TGCCTCACTG ATTAAGCATT CANCTGAGCG CCGGTCGCTA GCGTTTCGGT GGAGCAGACA ATTGTACTGA AGGGCCTCGT GGAAATGTGC ATANATGCTT ATCAGAGCAG PATAACCCTG SCGTCAGACC GCCGCCTTA GCGGATGCCG CACTITICGG TTCGTCTCGC

[Key to 10/15:]

Vektor = Vector

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ASSERVABERAD SUPPRESSION TO TOTAGO CONTINUE CANGINGTO CONGCINA COCCIGATO TOTAGADA CONTINUE TOTAGADA CONTINUE CONTINUE ACCIGATAC CONCIGARACIÓN CONGCICACO CONTINUE CONTINUE ACCIGATAC CONTINUE CONTINUE ACCIGATAC CONCIGARACIÓN CONGCICACO CONTINUE CONTINUE ACCIGATAC CONTINUE CONTINUE CONTINUE ACCIGATAC CONTINUE C

Senneral Lac-Promotor

Sequenza Lac-Operator

安徽部市第 Ribosomen-Bindungsstelle

Ampicillin-Resistenz-Gen

Marker puc Plasmid-Replications-Origin

Pati Ncoi Bargi EcoRi

Die Sequenz enthält eine silent mutation (Reconstates), die laut Sequenzanalyse eindeutig vorhanden ist:

Soll: AIT, Ist. ATH - Ilc; Codon usage (gesamtes Genom B. coli) indert sich von 30,3 zu 25,1 (Frequenz pro Tausend)

[Key to 11/15:]

Sequenz: Lac-Promotor = Sequence: Lac-promoter

Ribosomen-Bindungsstelle = Ribosome binding site

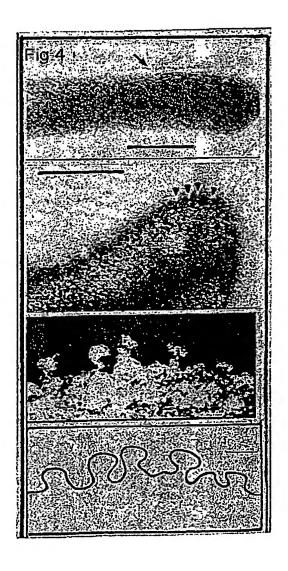
Ampicillin-Resistenz-Gen = Ampicillin resistance gene

Klonierungsstellen = Cloning Sites

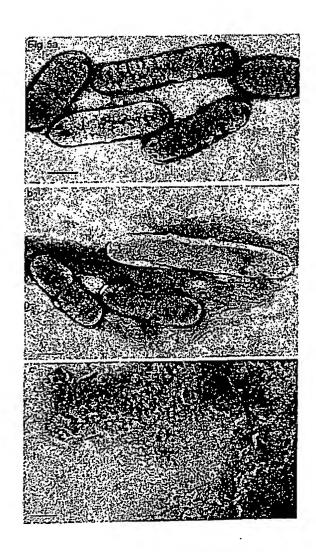
[last sentence:] The sequence contains a silent mutation (), which is clearly present according to the sequence analysis:

To be: ATT, Is: AT -> IIe; codon usage (entire E. coli genome) is changed from 30.3 to 25.1 (frequency per thousand)

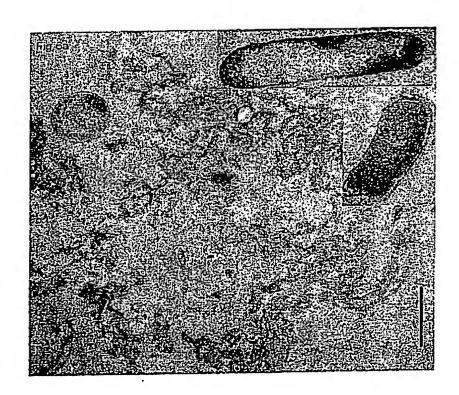
12/15



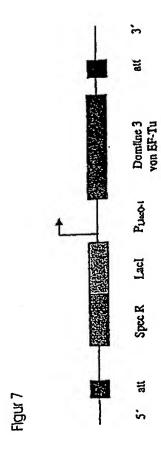
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[Key to Figure 7:]

Figur = Figure

Domäne 3 von EF-Tu = Domain 3 of EF-Tu

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